Larval and Early Benthic Stages of *Brachidontes granulata*

(Bivalvia : Mytilidae)

BY

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(2 Plates)

INTRODUCTION

Brachidontes granulata (Hanley, 1843) is a mytilid distributed from Lobos, Perú (6°S) to Isla Chiloé and Seno Reloncaví, Chile (43°S) (SOOT-RYEN, 1959). It may be found attached to undersides of rocks in the lower intertidal zone (MARINCOVICH, 1973), and forming small groups under the mats of various algae in intertidal pools in the rocky areas of Central Chile (STUARDO, 1960). In Montemar, Valparaíso (33°S), *B. granulata* is more abundant in the semisheltered zone. Other mytilid species that occur in the same area are: *Perumytilus purpuratus* (Lamarck, 1819), *Semimytilus algosus* (Gould, 1850) and *Aulacomya ater* (Molina, 1782).

Brachidontes granulata has been included in the subgenus or genus *Hormomya* together with *Mytilus adamsianus* Dunker, 1857 (SOOT-RYEN, 1955). However, KEEN (1971), adopting a conservative classification recognizes only the genus *Brachidontes*. This matter needs clarification in order to properly evaluate the differences found in the larval stages of the species included in *Brachidontes sensu lato*.

The following contributions have been made in relation to the larvae of bivalve species from the Chilean coast: RANSON (1960), HOLLIS & MILLAR (1963), PADILLA & ORREGO (1967), and SOLÍS (1967) on *Ostrea chilensis;* WALNE (1974) on *Choromytilus chorus* (misspelled as *C. choro*); PADILLA (1973) on *Mytilus edulis chilensis* and SOLÍS, SÁNCHEZ & NAVARETTE (1976) on bivalve larvae from the Estero de Castro. Only RANSON (*op. cit.*) and SOLÍS, SÁNCHEZ & NAVARETTE (*op. cit.*) described the morphology of the larval shell.

No descriptions of larvae of either *Hormomya* or *Brachidontes* are known to us, except the paper of CHANLEY (1970) on *B. recurvus* from the western North Atlantic Ocean. YOSHIDA (1937) described the larvae and early juveniles of *B. senhausi* (Reeve), but according to KURODA *et al.* (1971) this species belongs to the genus *Musculus*.

The purpose of this paper is to describe the larvae and early benthic stages of *Brachidontes granulata* so that they may be identified in planktonic and benthic samples. This information should prove especially useful in areas where other species of Mytilidae are cultivated for commercial purposes.

MATERIALS AND METHODS

Sexually mature adult mussels were collected at the intertidal zone near Montemar. In the laboratory all specimens were washed and placed in plastic dishes with filtered sea water (0.8 μm membrane filter) and held in an incubator at 6°C for 12 hours. Thereafter, they were changed to filtered sea water at 16°C where they usually began to spawn about 5 hours later.

Unfertilized eggs were poured through a set of nylon screens with mesh openings from 150 to 55 μm to remove debris. They were immediately placed in a finger bowl with filtered sea water, where some drops of sperm suspension were added. Fertilized eggs were kept without movement for 30 minutes, being transferred to a culture vessel with 2 L of filtered sea water at laboratory temperature (12 - 16°C). Three days later, when the end of the prodissoconch I was reached, air and a food mixture of *Tetraselmis* and *Nannochloris* were added; the concentration of algal food was not controlled. During the experiment the larvae were washed, sea water was changed and new food was added every 2 days. As the larvae of *Brachidontes granulata* were only cultured up to the early dissoconch stage, the description of the early benthic stages was made from recently settled individuals collected in the same area where adults came from.

Cleaned valves were examined, measured and photographed, using a Leitz Ortholux microscope with an Orthomat camera.

The terms prodissoconch I (= prod. I), prodissoconch II (= prod. II) and dissoconch follow WERNER (1939) and REES (1950). The terms veliger and pediveliger are defined in CHANLEY & ANDREWS (1971).

RESULTS

Brachidontes granulata is a dioecious species. Spawned eggs were brown in color with a mean diameter of 66.3 μm and a size range of 63 to 73 μm; sample size: n = 100.

The prod. I was colorless, transparent, and the measurements in μm were: mean length 117, s. d. 5.3, range 105 - 126; mean height 69.7, s. d. 3.9, range 63 - 73.5; n = 100.

The evidence of the prod. II development was a yellow edge in the shell material surrounding the prod. I. Simultaneously with the initial growth of the prod. II appeared a taxodont dentition, bearing an indeterminate number of small central teeth and 2 more prominent ones at the ends of the hinge line. As the larval shell was growing, the 2 largest teeth increased in size, and in number to 6 or 7, forming the curved end of the provinculum in the pediveliger stage. The umbo appeared at about 150 μm larval length as the result of the growing towards the central side of the largest provincular teeth. The dorsal side of the hinge line remained relatively flat, forming a broadly rounded umbo.

The eyespots appeared at 160 μm larval length. A notorious ligament slightly displaced towards the posterior side could be seen in larvae 180 μm in length. As larvae attained a length of 190 μm, the foot was evident between the valves and became functional at 200 μm length. The velum disappeared at 215 μm larval length.

At the pediveliger stage the larval shell of *Brachidontes granulata* could be characterized as follows: yellow prodissoconch, anterior margin curved, posterior end almost straight, ventral margin nearly circular, shoulders almost straight, the anterior being longer than the posterior one and not sloping as steeply, umbo broadly rounded and more prominent in width than in height. The mean length in μm of the prod. II was 215, s. d. 5.3, range 200 - 231; n = 100.

The correlation of the major features of larval development and sizes of *Brachidontes granulata* reared in the laboratory, is summarized in Table 1, and the main characteristics of the larval shell are shown in Figures 1 to 6.

Table 1

Summary of the major features of larval development in *Brachidontes granulata*. Temp. 12-16°C; S.34-35‰

Stage or distinctive feature	Mean size microns	Mean age
Unfertilized egg	66.3	
Trochophore	73.0	20 hours
Prodissoconch I	117.0	3 days
Early umbo	150.0	12 days
Appearance of eyespots	160.0	22 days
Ligament evident	180.0	28 days
Foot	190.0	30 days
Functional foot (pediveliger)	200.0	40 days
Loss of velum	215.0	55 days

The shell of the early juveniles collected from the intertidal zone shows the following characteristics (Figures 7 to 13): The dissoconch is colorless and transparent; this feature serves to easily distinguish the limit of the yellow prod. II. About 290 μm in length (maximal distance parallel to the hinge), the shell shows a conspicuous growth towards the ventero-posterior end. At a length of about 400 μm, the anterior ventral margin of the shell is straight and the umbo is prominent; also the first disso-

Explanation of Figures 1 to 7

Larval and early benthic stages of *Brachidontes granulata*

Figure 1: Internal view of the larval shell, right valve; length 126 μm

Figure 2: Internal view of the larval shell, right valve; length 190 μm

Figure 3: Internal view of the larval shell at the pediveliger stage, right valve; length 210 μm

Figure 4: Hinge structure of the larval shell at the pediveliger stage, dorsal view. Shell length 210 μm

Figure 5: Hinge structure of the larval shell at the pediveliger stage, internal view. Shell length 210 μm

Figure 6: Larva at late pediveliger stage; length 220 μm

Figure 7: Internal view of the right valve at early benthic stage showing the initial dissoconch; length 240 μm


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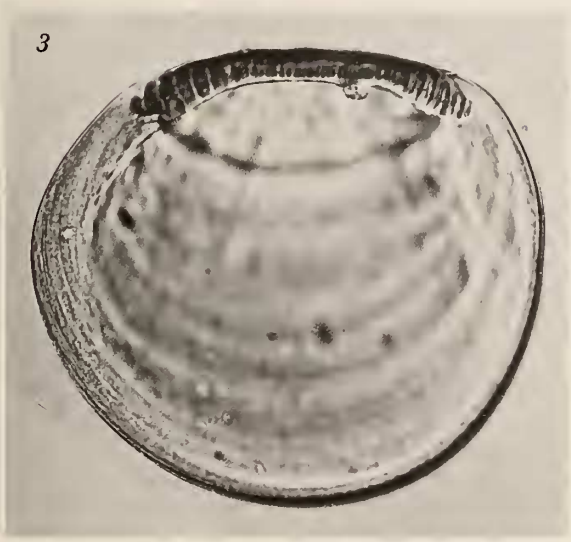
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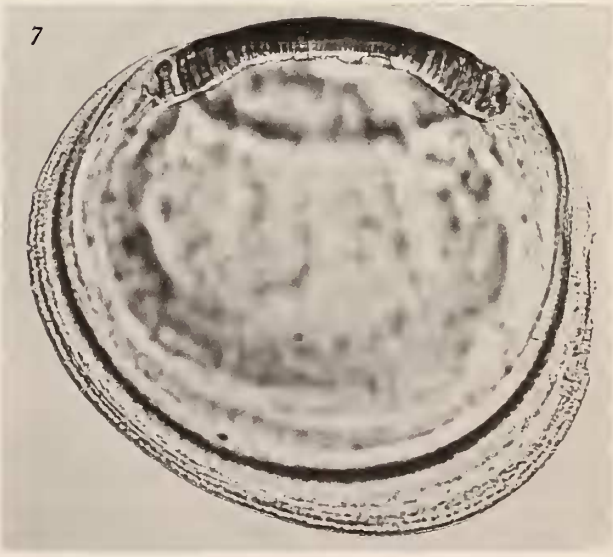
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conch teeth (Cox, 1969) develop along the posterodorsal margin some distance beyond the larval hinge. At shell lengths ranging from 450 to 550 μm, a new zone grows in the dissoconch; this new zone is characterized by the presence of radial ribs deeply impressed, a slight sinus in the ventral margin and a white coloration contrasting with the first colorless zone of the dissoconch.

DISCUSSION

Although the main purpose of this work was the description of larval and post-larval stages of *Brachidontes granulata,* the method used allowed us to obtain additional information on the development of the larvae under laboratory conditions.

The common methods to induce spawning of sexually mature bivalves in the laboratory are thermal, chemical and electrical shock, sex products stimulation and stretching the posterior adductor muscle (Loosanoff & Davis, 1963). The review of the literature reveals the difficulties of inducing bivalves to spawn, and that there is no agreement as to which stimulus is best for each species. The insertion of a small wooden wedge between the shells and stretching of the adductor muscle was unsuccessful in *Brachidontes granulata.* However, rapid cooling and warming of the water always stimulated spawning of abundant eggs and sperm, giving rise to healthy larvae.

From the investigations carried out to date (Carriker, 1961; Loosanoff & Davis, 1963; Bayne, 1965; Loosanoff, Davis & Chanley, 1966; DeSchweinitz & Lutz, 1976), it is reasonable to infer that in bivalves many features, such as larval shells. eyespots, foot, etc., appearing during the larval stages up to metamorphosis, seem to be primarily dependent on size rather than age, temperature, food, or other environmental factors. This conclusion suggests the validity of correlating the larval dimensions with the shape of the larval shells, as well as the other structures already mentioned, even if the individuals may show small variations in the size at which a given feature appears. This generalization has led us to rear *Brachidontes granulata* with minimal laboratory requirements, regardless of the duration of the larval development.

Cultured larvae reached the pediveliger stage by the 40th day at a mean size of 200 μm in length. Later, the velum gradually degenerated, disappearing by the 55th day. These larvae were kept alive until the end of the experiment for another 18 days, and no attachment was observed during this time; this fact suggests a delay of metamorphosis in *Brachidontes granulata* larvae. Our results agree with those of Bayne (1965) on *Mytilus edulis,* mainly in relation to the events that occur during the delay of metamorphosis. This same author emphasizes that the major factor influencing the delay of metamorphosis is the availability of a suitable substrate capable of stimulating the secretion of the byssus gland. Although no experiments on the effects of environmental factors in the metamorphosis were carried out by us, the fact that suitable substrates were not offered to pediveligers, might have been the cause of non-attachment in *B. granulata* larvae.

From the studies of Loosanoff (1961), Carriker (1961), Loosanoff & Davis (1963) and Bayne (1965), we infer that it is difficult to ascertain the precise moment of metamorphosis. It is rather a gradual process, preceded by the pediveliger stage, and ending with the development of a functional byssus gland and the appearance of the dissoconch shell.

Neither functional byssal gland nor growth of the shell were observed during the 33 days after the larvae of *Brachidontes granulata* reached the pediveliger stage, except for a narrow white band of shell that marked the beginning of the dissoconch.

While culturing *Anomia simplex,* Loosanoff (1961) observed the disappearance of the velum, retention of a functional foot, and the beginning of the dissoconch without attachment to the substratum. This experience is very similar to ours, except that the shell of *Brachidontes granulata* did not grow appreciably as in *A. simplex.* Loosanoff (*op. cit.*) named this phenomenon 'partial metamorphosis without attachment to the substratum.' We do not know if the byssal gland in *B. granulata* could become functional in case that a suitable substrate had been added after the beginning of the dissoconch growth. These "abnormalities" in the metamorphosis process should be further investigated before any feature delimiting the end of metamorphosis can be properly ascertained.

Larvae of *Brachidontes granulata* are typical pelagic mytilid larvae. Compared with those of *B. recurvus* from the coast of Virginia in the North Atlantic (Chanley, 1970), the former have a more rounded shell, the large teeth of the provinculum appear at a bigger larval size (126 against 105 μm in *B. recurvus*) and have a distinct ligament. Egg diameter, umbo shape and size at which the eyespots appear are about the same in both species.

A shell area located immediately beyond the prodissoconch and differing conspicuously from the final dissoconch in the early bottom stages of some mytilids, has been described as an "interdissoconch" (Jørgensen, 1946; Rees, 1950).